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Assessment of the Immune Responsiveness of Mice Irradiated with Continuous Wave or Pulse-Modulated 425 MHz Radio Frequency Radiation

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16. ABSTRACT

Groups of female BALB/C mice were irradiated with 425-MHz radio frequency (RF) radiation either continous wave (CW) or pulse modulated (PM, 1-ms pulse width, 250 pulses/s). Mice were irradiated in a rectangular strip-transmission line at average forward powers of 78, 17.7, or 5 W for CW and 17.7, 5, or 1.25 W for PM. The mean specific absorption rate , as measured using twin-well calorimetry was 7.7 W/kg for a forward power of 70 W. No differences in the mitogen-stimulated response of lymphocytes or in the primary antibody response to sheep erythrocytes or polyvinylpyrrolidone were observed between irradiated and sham-irradiated mice, nor between mice exposed to either CW or PM 425-MHz RF radiation.

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Brief Communication

Assessment of the Immune Responsiveness of Mice Irradiated With Continuous Wave or Pulse-Modulated 425-MHz Radio Frequency Radiation

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Groups of female BALB/C mice were irradiated with 425-MHz radio frequency (RF) radiation either continuous wave (CW) or pulse modulated (PM, 1-ms pulse width, 250 pulses/s). Mice were irradiated in a rectangular strip-transmission line at average forward powers of 78, 17.7, or 5 W for CW and 17.7, 5, or 1.25 W for PM. The mean specific absorption rate, as measured using twin-well calorimetry was 7.7 W/kg for a forward power of 70 W. No differences in the mitogen-stimulated response of lymphocytes or in the primary antibody response to sheep erythrocytes or polyvinylpyrrolidone were observed between irradiated and sham-irradiated mice, nor between mice exposed to either CW or PM 425-MHz RF radiation.

Key words: 425 MHz, mice, continuous wave, pulse modulated, immune response, lymphocytes

There are reports of quantitative differences in the biological effects produced by exposure of experimental animals to pulse-modulated (PM) compared with continuous wave (CW) RF radiation [Baranski and Edelwejn, 1975; Czerski, 1975; and Frey et al, 1975]. If such differences exist, then there is the potential for additional complexity in setting RF exposure standards for humans (ie, standards based on average power only). In an attempt to determine if the immune system would be differentially affected by CW versus PM RF radiation, we exposed mice to 425 MHz under either mode. Our decision to use 425-MHz was based on previous studies where effects on lymphocytes were observed [Smialowicz et al, 1982]. At the time of these experiments we were also aware that a new radar system, ie, Precision Acquisition of Vehicle Entry-Phased Array Warning System (PAVE PAWS) was being deployed by the U.S. Air Force. These radars operate

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within a frequency range of 420- to 450-MHz, using pulse modulation with a 0.25 duty cycle, maximum. Consequently, we chose a similar duty cycle for our experiments.

Groups of four female BALB/C mice (8–10 weeks of age, Charles River Labs, Willmington, MA) were confined individually in small containers 6 cm on a side, consisting of 1.5-mm-thick clear polycarbonate with a polyethylene top. The sides and top of the containers had 6–8 ventilation holes 5 mm in diameter. The mice were exposed to 425-MHz RF radiation in a rectangular strip-transmission line described by Smialowicz et al [1982]. Two containers, each holding one mouse, were placed on either side of the center conductor. An equal number of sham-irradiated control mice were placed in a foamed polystyrene chamber adjacent to the transmission line. The temperature in the sham chamber and the transmission line was maintained at 22 ± 0.5 °C with an air flow of 1.3 m^3/min. Humidity was not controlled.

Pulse-modulated RF power having the approximate characteristics of the PAVE PAWS radar (ie, 1-ms pulse width and pulse repetition frequency (PRF) of 250 pulses/s) was supplied by a RF power generator (AIL 125 AILTECH, Inc, Farmingdale, NY) that was modulated by a pulse generator (Model 6613, Texas Instruments, Dallas, TX). Two or three groups of four mice each were irradiated for 1 h on each of 5 consecutive days using continuous wave 425-MHz at forward powers of 78, 17.7, or 5 W. For pulse modulation, the irradiated mice were exposed at average forward powers of 17.7, 5, or 1.25 W with peak forward powers of 70.8, 20, and 5 W, respectively. Sham-irradiated controls were always run simultaneously with irradiated mice.

The average specific absorption rate (SAR) of each of the mice in the four positions irradiated at a forward power of 70 W (CW) was determined by twin-well calorimetry [Kinn, 1977]. No statistically significant difference was observed in SAR values for mice in the four different positions with the other three occupied by mice, within the transmission line. The average SAR for the group of four mice was 7.7 ± 1.8 W/kg (± SD) under these conditions.

Immediately following the fifth and final irradiation session, mice were immunized with sheep red blood cells (SRBC) and 4 days later the primary immune response was determined using the direct plaque-forming cell (PFC) assay [Smialowicz et al, 1979].

Table 1 shows the primary immune response to SRBC of irradiated mice. No difference in the immune response was observed for mice irradiated with either CW or pulse-modulated 425-MHz radiation. Multivariate analysis of variance methods were used to analyze the data.

Other mice were irradiated under similar conditions with CW or PM 425-MHz and bled from the orbital sinus immediately following the last irradiation. Blood smears were prepared on slides with Wright's stain, and 200 leukocytes were differentially counted for lymphocytes, monocytes, eosinophils, and polymorphonuclear (PMN) leukocytes. Erythrocyte (RBC) count, leukocyte (WBC) count, hematocrit (Hct), and hemoglobin (Hgb) concentration in the blood were evaluated on a Coulter counter (Model ZBI-6 Coulter Electronics, Inc., Hialeah, FL). Table 2 shows the hemograms for mice irradiated at a mean forward power of 78 W CW (SAR = 8.6 W/kg) and 17.7 W PM (SAR = 1.9 W/kg). A significant increase ($P<.05$, analysis of variance) in the leukocyte count and absolute lymphocyte count was observed for mice irradiated at 78 W CW compared with their respective sham controls. No difference was observed in the hemograms of mice irradiated at 17.7 W PM compared with controls. A comparison of the hemograms between mice irradiated at 78-W CW and 17.7 W PM by multivariate analysis of variance revealed no difference between the two groups. It appears that the observed differences

are due to spuriously low counts for the control mice as opposed to an effect of RF radiation on peripheral blood lymphocytes; however, this requires further investigation.

In other experiments mice were irradiated with 425-MHz CW or PM radiation at a forward power of 17.7 for 1 h on 5 consecutive days. Immediately following the last irradiation, mice were immunized with a single intravenous injection of 0.125 μg polyvinylpyrrolidone (PVP). Five days later the primary immune response was determined using the direct PFC assay with PVP-coated SRBC as described by Lake and Reed [1976]. No difference in the response to PVP by mice irradiated with either CW or PM RF radiation or between control and irradiated mice was observed.

The effect on the in vitro mitogen-stimulated response of lymphocytes from mice irradiated with CW or PM modulation RF radiation was also examined. Again, groups of mice were irradiated for 1 h on 5 consecutive days to either CW or PM at a mean forward power of 17.7 W. Twenty-four hours following the last irradiation, the mice were killed and spleen cell suspensions were prepared for the mitogen response assay [Smialowicz et al, 1979]. Lymphocyte responsiveness was determined for the following mitogens at several different concentrations: phytohemmagglutinin (PHA, Burroughs Wellcome Co, RTP, NC); concanavalin A (ConA, Miles Lab, Kankakee, IL); Escherichia coli 0.28: B12 lipopolysaccharide (LPS, Difco Lab, Detroit, MI), and pokeweed mitogen

TABLE 1. Primary Immune Response to Sheep Red Blood Cells in Mice Irradiated With 425-MHz, CW, or PM

Mean forward power (W)		Mean log PFC/spleen ± SE[a]			
		Continuous wave		Pulse modulated	
		(Control)	(Irradiated)	(Control)	(Irradiated)
78	(8.6)[c]	5.60 ± 0.04	5.64 ± 0.03	ND[b]	ND
17.7	(1.9)	5.58 ± 0.03	5.59 ± 0.05	5.58 ± 0.04	5.60 ± 0.04
5	(0.5)	5.61 ± 0.03	5.58 ± 0.04	5.58 ± 0.05	5.59 ± 0.04
1.25	(0.1)	ND	ND	5.66 ± 0.02	5.58 ± 0.04

[a]N = 8–12 mice for each datum point.
[b]Not done.
[c]SAR in W/kg.

TABLE 2. Peripheral Blood Counts of Mice Irradiated With 425-MHz, CW, or PM

Parameter	Continuous wave (78 W)[a]		Pulse Modulated (17.7 W)	
	Control(12)[b]	Irradiated(12)	Control(12)	Irradiated(12)
RBC (10^6/mm^3)	9.94 ± 0.38	10.0 ± 0.42	10.1 ± 0.68	10.0 ± 0.44
WBC (10^3/mm^3)	3.84 ± 0.81	4.69 ± 0.97[c]	4.93 ± 1.34	4.92 ± 1.40
HgB (g/dl)	16.5 ± 0.73	17.0 ± 0.72	17.1 ± 1.34	16.8 ± 0.87
Hct (ml%)	49.1 ± 2.55	48.4 ± 3.10	50.1 ± 3.76	50.5 ± 2.48
PMN (10^3/mm^3)	0.57 ± 0.19	0.61 ± 0.25	0.67 ± 0.37	0.73 ± 0.27
Lymphocytes (10^3/mm^3)	3.21 ± 0.68	4.02 ± 0.83[c]	4.19 ± 1.02	4.09 ± 1.25

[a]Mean forward power.
[b]Number of mice, $\bar{X}$ ± SD.
[c]$P < .05$, analysis of variance.

(PWM, Grand Island Biological Co, Grand Island, NY). No difference was observed in the responsiveness of lymphocytes from CW- or PM-irradiated mice nor between control and irradiated mice for all of the mitogens tested.

In previous work [Smialowicz et al, 1982], we observed a potentiation of the mitogen responsiveness of lymphocytes from rats irradiated pre- and postnatally with 425-MHz CW at a forward power of 20 W. In the present study no difference was observed for lymphocytes from mice exposed to CW or PM. The difference in exposure parameters and in species between the previous work and the present study may account for our failure to observe an effect.

Examination of the effect of CW and PM RF radiation on the primary antibody response to the T- (thymus-derived) lymphocyte-dependent antigen SRBC and the T-independent antigen PVP also failed to show any changes. The observed difference in leukocyte and lymphocyte counts of mice exposed to 425-MHz CW radiation at a mean forward power of 78 W is apparently due to spuriously low counts in the sham control mice, although this interpretation requires further work.

The failure to detect a change in the immune response of mice irradiated under either CW or PM conditions does not directly answer the question as to whether or not a qualitative difference in biological effectiveness exists between these two modes. This work does, however, demonstrate that either modality, under the conditions described, is of no consequence to the integrity of the immune system functions of the mice examined in this study.

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DISCLAIMER

This paper has been reviewed by the Health Effects Research Laboratory, U.S. Environmental Protection Agency, and approved for publication. Mention of trade names or commercial products does not constitute endorsement or recommendation for use.

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